THE INFLUENCE OF THE ENVIRONMENT ON THE SIZE OF EXPECTED CLASSES.

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In crosses in which rudimentary wings are involved, it has been apparent, since this race was first bred, that the classes containing rudimentary wings often run far behind expectation. The experiments made clear that the character rudimentary wings is a Mendelian recessive and is sex-linked. The deficiencies that appeared were assigned to viability of these flies. We have found meanwhile for other stocks that by breeding *pairs* of flies in large bottles, with an abundance of food, kept in good condition, there was a very marked increase in number of those classes that are deficient in number if many flies are bred in small bottles, or even in large bottles if so many parents are used that crowding of the larvae takes place. It was determined, therefore, to repeat the experiments with rudimentary wings under the most favorable conditions that our experience had made known to us. In order to avoid the possible criticism that the stock might have changed, a control culture *en masse* was again made in which crowding took place.

Results similar to these with rudimentary wings had also come up in crosses in which a new mutant, "strap wing" was involved. This factor is not sex linked, but belongs to our second group. Similar experiments were carried out with this stock.

THE VIABILITY OF THE RUDIMENTARY WINGED RACE.

In a paper in *Science*, 1911,[1] an F_2 count is given in which 5,850 long-winged flies (♂ and ♀) and 83 rudimentary-winged males are recorded. The expectation is that of these 5,850 flies one third should be long-winged males, or 1,950. This number is also the expectation for the rudimentary-winged males. Instead of 1,950 there are only 83 males or 1/23 the expected number.

[1] *Science*, Vol. XXXIII., March 31, 1911.

The F_1 generation of the reciprocal cross was published in *Science*, 1912.[1] The rudimentary-winged female bred to a long-winged male gave 381 long-winged daughters and only 3 rudimentary-winged sons where equality was expected. Whether the lack of sons here is due entirely to viability, or to other conditions as well cannot be stated.

In a very brief paper in 1911[2] some other data were given that showed the rudimentary classes running behind expectation. These data were corrected and expanded in another paper,[3] to which reference may now be made. An F_2 count is given there, that is an extension of the data published in the first paper referred to above. There are 14,309 long-winged grandchildren (♂♂ and ♀♀) and 115 males with rudimentary wings. The expectation here is that one third of 14,309 or 4,769 flies should have rudimentary wings. The entire number is 115, or only 1/41 of the expected number.

There is an F_1 count of the reciprocal cross when 68 long-winged daughters and 3 rudimentary-winged sons appear. This ratio is not lower than that given above, and may safely be ascribed to viability. In the F_2 count of the same combination the following classes and numbers were realized:

Long ♀	721
Long ♂	698
Rud. ♀	163
Rud. ♂	237

The expectation is for equal numbers. The rudimentary males, while far behind expectation, are not so far behind as in other crosses cited, which is due to better treatment. This same statement applies to the remaining data of the 1912 paper for, at that time realizing more fully the influence of the environment on viability larger bottles with more food were used. Since most of the data involves miniature wings it is not cited here.

In the new experiments rudimentary-winged males were bred to long-winged (wild) females; and long-winged daughters and sons obtained. The daughters were then back-crossed to rudimentary males either in pairs or *en masse*. When bred *en masse*

[1] *Science*, Vol. XXXIV., March 22, 1912.

[2] *Proc. Soc. Exp. Biol. and Medicine*, VIII., February, 1911.

[3] *Zeit. f. induktive Abstammungs und Vererbunglehre*, VII., 1912.

the following offspring were obtained. The expectation is for equality in all four classes:

Long ♀	527
Long ♂	489
Rudimentary ♀	7
Rudimentary ♂	31

There is an enormous deficit in the classes of rudimentary flies. Instead of equality there are only 1/27 as many as expected.

When on the other hand the F_1 females were back-crossed *in pairs* the following totals were obtained:

Long ♀	1,717
Long ♂	1,545
Rudimentary ♀	1,120
Rudimentary ♂	1,179

There is an approach to equality in the last case (1 : .7), and, in consequence, the contrast with the preceding data is striking. For purposes of more detailed comparison the data for the two mass cultures and for the 20 pairs is given:

TABLE I.

BACK-CROSS PAIRS.

No.	Long ♀.	Long ♂.	Rudimentary ♀.	Rudimentary ♂.
7	133	117	94	102
8	59	43	55	47
9	107	114	67	66
10	70	55	46	64
11	62	60	53	36
12	49	36	51	41
13	86	76	31	43
14	135	101	61	104
15	107	102	61	79
16	67	44	60	54
17	116	107	73	70
18	92	80	72	68
19	87	77	84	53
20	16	14	12	6
21	78	101	73	72
22	83	60	44	50
23	88	81	62	91
24	52	49	35	37
25	95	100	56	65
26	135	128	30	31
Total.....	1,717	1,545	1,120	1,179

BACK-CROSS EN MASSE.

28	248	235	4	14
29	279	254	3	17
Total.....	527	489	7	31

This same mating was again made both in mass and in pairs. The expectation is equality of long and rudimentary wings. The mass cultures gave:

Long ♀ 341
Long ♂ 337
Rudimentary ♀ 51
Rudimentary ♂ 64

When the experiment was made with pairs the following totals were obtained, (with a ratio of 1. : .63):

Long ♀ 1,676
Long ♂ 1,431
Rudimentary ♀ 930
Rudimentary ♂ 1,022

For detailed comparison the counts of the two mass cultures and of the 29 pairs taken separately are given in Table II.

TABLE II.

BACK-CROSS PAIRS.

No.	Long ♀.	Long ♂.	Rudimentary ♀.	Rudimentary ♂.
31	161	122	108	154
32	15	7	5	4
33	154	121	27	30
34	82	103	38	66
35	103	81	11	13
36	143	74	12	42
37	86	84	62	49
38	59	79	51	57
39	123	98	67	61
40	164	142	70	85
41	63	59	71	54
42	72	45	47	42
43	56	65	55	55
44	47	51	47	46
45	45	37	34	25
46	93	64	42	47
47	45	50	46	33
48	63	47	44	55
49	59	45	52	65
50	63	57	41	39
Total.....	1,676	1,431	930	1,022

BACK-CROSS EN MASSE.

51	154	135	18	19
52	187	202	33	45
Total.....	341	337	51	64

Since rudimentary females are sterile with rudimentary males the stock of rudimentary wings is maintained by breeding rudimentary males to heterozygous, long-winged females.

THE VIABILITY OF THE STRAP-WING RACE.

The peculiarities of structure and inheritance of this mutant will be described in another place. The stock contained "beading" and overlaps in appearance the vestigial wing. The "beaded" flies that appear in F_2 are classified with "normal" or long wing, those with vestigial-like wings are counted in with "strap." Strap bred to wild stock gives sons and daughters with long wings. These bred *en masse* during August gave the following results in eleven cultures. About six to ten F_1 flies were used in each culture in a medium sized bottle, and the food conditions were made as favorable as possible.

	Normal.	Strap.	Ratio.
1.	224	59	1 : 3.8
2.	236	63	1 : 3.7
3.	471 (+39 curved)	28	1 : 17.0
4.	216	45	1 : 4.8
5.	103	11	1 : 9.3
6.	277	24	1 : 11.5
7.	162	14	1 : 11.6
8.	464	23	1 : 20.1
9.	494	82	1 : 6.2
10.	288	44	1 : 6.5
11.	164	34	1 : 4.8
Total.....	3,099 (+39)	427	1 : 7.2

The ratios range from 1 : 3.7, to 1 : 20.1. In order to compare these results with matings in pairs, fifteen pairs were mated. After ten days the parents were placed in a new bottle with fresh food and in some cases they were carried to a third bottle. The results are given in the next table (III.).

The results from pairs approximate more nearly to expectation (1 : 3); the totals give a ratio of 1 : 3.57. The individual lots

TABLE III.

F_2 FROM STRAP ♂ × WILD ♀.

No.	Brood I.					Brood II.					Brood III.				
	Normal.		Strap.		Ratio to 1.	Normal.		Strap.		Ratio to 1.	Normal.		Strap.		Ratio to 1.
	♀	♂	♀	♂		♀	♂	♀	♂		♀	♂	♀	♂	
1	97	89	27	17	4.2	218	199	60	47	3.9	—	—	—	—	—
2	112	116	24	20	5.2	148	141	23	26	5.9	48	44	16	14	3.1
3	90	76	13	12	6.6	88	88	8	5	13.5	—	—	—	—	—
4	117	101	37	14	4.3	99	130	38	28	3.5	25	41	10	16	2.4
5	73	63	24	31	2.5	80	88	34	30	2.6	7	3	1	1	—
6	79	90	23	26	3.5	119	100	30	24	4.1	10	14	2	5	—
7	105	86	31	27	3.3	121	115	27	37	3.7	50	59	12	23	3.1
8	94	91	32	22	3.4	120	113	39	36	3.1	24	37	10	6	3.8
9	95	87	19	31	3.6	101	106	29	29	3.6	59	42	17	27	2.3
10	72	60	31	26	2.3	46	59	22	20	2.5	5	—	1	—	—
11	66	63	12	22	3.8	57	36	5	7	7.7	—	—	—	—	—
12	79	68	27	38	2.3	77	77	28	24	3.0	3	3	1	—	—
13	79	65	26	15	3.5	54	62	17	18	3.3	37	29	10	5	4.4
14	85	80	28	26	3.1	78	90	22	21	3.9	32	34	11	7	3.8
15	62	61	14	15	4.1	63	53	21	26	2.5	30	17	7	7	3.4
	1,305	1,196	368	342	3.52	1,479	1,457	403	378	3.75	330	323	98	111	3.12

are worthy of inspection. The ratios run more evenly and range from 1 : 2.3 to 1 : 7.7 with one exceptionally high at 1 : 13.5. The counts from day to day (not recorded here) show that the F_2 strap-winged flies hatch later than the flies with normal wings, so that as the cultures run out, the relative number of strap-wing flies increases, and unless the cultures are exhausted when the count stops there will be a deficiency of strap-wing flies. An attempt was made to run the cultures to a finish, although this cannot always be done. If the cultures dry up, a disproportionately large number of strap-winged flies will be destroyed, and this will account in part for the deficiency in this class. These results are not affected by the age of the parents, as shown by comparison of the ratios in the totals for the second (1 : 3.75) and third (1 : 3.12) broods with that of the first (1 : 3.52).

The above pairs were mated in November and December, 1913. In order to make a more exact comparison, a few mass cultures were made in December from the same stocks. The results are shown in the next table (IV.).

In addition to the preceding data there were other crosses of strap male by wild female that had been made nine months

TABLE IV.

No.	Normal.	Strap.	Ratio.
1	455	67	1 : 6.8
2	365	65	1 : 5.6
3	330	75	1 : 4.4
Total.....	1,150	207	1 : 5.5

before those recorded above. In these earlier experiments in mass cultures no attempt was made to avoid crowding through use of too many parents, although the bottles were otherwise maintained in good condition. The following F_2 counts were made:

	Long ♀ and ♂.	Strap ♀ and ♂.	Ratio.
1.	739	76	1 : 9.7
2.	411	55	1 : 7.5
3.	247	22	1 : 11.2
4.	410	59	1 : 6.9
5.	464	54	1 : 8.6
6.	266	33	1 : 8.0
7.	490	61	1 : 7.8
8.	342	43	1 : 8.1
9.	137	13	1 : 10.5
10.	493	3	1 : 164.0
11.	245	8	1 : 30.6
12.	197	0	———
13.	818	137	1 : 6.0
14.	614	125	1 : 4.9
Total.....	5,873	689	1 : 8.53

Whether the two very high ratios under 10 and 12 should be included may be questioned. It is possible though not probable that contamination took place. Excluding these the ratios vary from 1 : 4.9 to 1 : 30.6.

CONCLUSIONS.

The experiments show that under unfavorable conditions due to crowding the mutant forms, rudimentary wings and strap wings, run behind expectation. That the result is due to crowding is shown when cultures of sister individuals are made *in pairs* with abundant room and food. There is then a closer, and in some cultures a complete agreement with expectation. In experiments in which it is only necessary to show whether a character is recessive or dominant, and whether it is, or is not

linked to other characters these deficient ratios, while unfortunate, present no serious difficulties, but in experiments in which it is necessary to determine accurately the linkage ratios the difference becomes serious.

The rudimentary and strap-winged mutants have been used to test the effects of crowding due to mass breeding, because they have been found to show such effects more than any other stocks. In the case of other mutants that give, even in mass cultures, nearly the theoretical values, the method which we now generally employ of breeding by pairs in large culture bottles with plenty of food eliminates almost entirely disturbances due to viability.